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The work was divided into an elementary and an advanced course, the former for those who took up the study of botany for the first time, and the latter for those who had already made some progress in the study. The attendance was large, considerably exceeding one hundred, and was composed almost entirely of teachers of maturer years, in all departments of school work, from the kindergarten to the high-school and academy.—CHARLES E. BESSEY.

VEGETABLE PHYSIOLOGY.¹

Fischer on Bacteria.—Under the title *Untersuchungen ueber Bakterien*, Dr. Albert Fischer contributes an important paper to a recent number of Pringsheim's *Jahrbücher für wissenschaftliche Botanik* (Bd. 27, H. 1, pp. 163, T. 5, Berlin, 1895). This paper consists of four parts: (1) New observations on the plasmolysis of bacteria; (2) The physiology of the flagella and of the movement; (3) The morphology of the flagella; (4) Classification. Of the five plates illustrating flagella, four are lithographic, and one is a collotype. The author appears to have made out pretty clearly for a good many forms that the contents of the bacterial cell is plasmolyzed even by a slight concentration of culture media such as takes place on the cover glass in drying or in the transfer of the organisms from a weaker to a more concentrated culture medium. This plasmolysis can be avoided by diluting the fluid very plentifully with water before making cover glass preparations from it. Only a very slight amount of sodium chloride is necessary to produce plasmolysis of a cover glass preparation, especially at the edge of the drop, viz.: 0.01 to 0.05 per cent. The occurrence of this phenomenon can be observed in a hanging drop as it dries. Plasmolysis disappears when watery stains are used, but is beautifully preserved by alcoholic stains, Ziehl's carbol fuchsin, or Delafield's haematoxylin. Many false conclusions have been drawn from such plasmolyzed bacteria. Here belong De Toni and Trevisan's genera *Pasteurella* and *Dicoccia*; the staining phenomena of the cholera vibrio, described by Rahmer; the bamboo-like joints sometimes seen in the anthrax bacillus; the polar bodies in the typhoid bacillus; the various granular structures in the tubercle bacillus, etc. The unstained, empty places

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in plasmolyzed bacteria have often been mistaken for spores. In weak salt solutions the phenomena of plasmolysis disappears in an hour or two; in strong solutions it disappears much sooner. This disappearance of plasmolysis and the reappearance of motility bear no relation to each other, but depend upon entirely different causes. To obtain good plasmolyzed cover-glass preparations that will fix and stain in that condition, the author recommends putting a trace of bacteria into a drop of a weak salt solution (0.25 to 0.50 per cent NaCl or 0.5 to 1.0 per cent KNO_3) and then carefully spreading out the drop so that it will dry in 3 to 10 minutes. The bacterial cell consists of a membrane, a protoplast in the form of a wall covering, and of cell sap, and has, consequently, the same structure as any other plant cell. Cell nuclei are still to be sought; a "centralkörper" is never present, when there seems to be one it is a misinterpretation due to the contracted protoplast, as in case of Bütschli's observations on *Spirillum undula*. In weak salt solutions which cause distinct plasmolysis (2.5 per cent KNO_3 ; 1.25 per cent NaCl, etc.) motile bacteria continue to move, often for hours. In stronger solutions (5–10 per cent KNO_3 , etc.), the movement ceases in a few minutes owing to the benumbing of the flagella, which, however, are never drawn back into the body of the bacillus, being in this respect quite like the motile organs of the Flagellata and unlike pseudopodia. In salt solutions which do not inhibit growth, but are strong enough to produce rigidity of the flagella, these organs continue to be produced. The same is true when 0.1 per cent carbolic acid or picric acid is added. Motility reappears when these inhibitory substances are removed. As in the flagella of the Flagellata the cilia of the Infusoria, and the lashes of ciliated epithelium the movement of the flagella in the bacteria is not independent of the protoplast, but nevertheless continues when the latter is disturbed by plasmolysis. Apparently, as in case of crushed infusoria a small fragment of the protoplast remaining attached to the base of the flagellum is sufficient to continue the movement. Rigidity of the flagella can be brought about in various ways—lack of oxygen, acid reactions, too much salt, mal nutrition, or the addition of poisons. On removal of these injurious influences the motility returns. In case on non-motile cultures of the hay bacillus the addition of $\frac{1}{2}$ per cent asparagin sufficed to induce motility quickly. In the making of cover-glass preparations various changes may take place in the flagella, they may be thrown off, or inrolled, or become swollen so as to be unstainable and unrecognizable. The inrolled flagella never unroll. They often appear as little foamy heaps of rings around the bacteria (typhoid bacil-

lus, hay bacillus, etc.) When the bacillus dies the flagella loose their power of swelling. The flagella often remain till the last, i. e., after the membrane and contents of the bacillus has disappeared. This ready swelling which is always at right angles to the long axis, makes the flagella in stained preparations always thicker than natural. The sprouting of the flagella from the body of the cell and their subsequent increase to full length consumes sufficient time so that its phases can be fixed and studied. In *Spirillum undula* it takes place before completed cell-division and from that end of the cell previously destitute of flagella. Continued cultivation in strong salt solutions, e. g. 4 to 5 per cent NH_4Cl , prevents motility, but does not interfere with the formation of the flagella. By movements of neighboring bacilli the flagella are often twisted into strands which are sometimes very large.

In *Bacillus subtilis* the spore is generally found in non-flagellate rods forming the pellicle, rarely in free swimming flagellate rods. The flagella of bacteria are not drawn back into the cell during spore formation. Involution forms of *Bacillus subtilis* bear no flagella, but in the involution forms of some other bacteria they are not thrown off. All motile bacteria possess flagella, and these are the sole organs of movement. Flagella are polar or diffuse according as they are restricted to one end of the cell or occur on any part of it. Polar flagella vary in number from one to several, and this number is characteristic for different species, except when the cells are dividing polar flagella are always at one end. The flagella of the bacteria are neither threads of protoplasm which can be thrust out and drawn back, nor dead appendages of the membrane moved by the protoplast. The substance of the flagellum possesses a life of its own, and the power of swelling and self-contraction. With the protoplast, of which they are a part, the flagella appear to be only loosely connected, yet the little protoplasmic remnant which in plasmolysis often remains attached to the base of the flagellum, and sometimes connects it with the shrunken protoplast is certainly to be regarded as a sign of such morphological union. In connection with the physiological diagnosis of the bacteria a morphological basis for classification is to be sought, and this the author thinks he has found for the rod-shaped bacteria in the number and position of the flagella and the shape of the spore-bearing cells. The author's classification is probably a step in the right direction, and will certainly lead to renewed efforts to determine the number and position of the flagella on a great variety of microorganisms, but, in the present state of our ignorance, it cannot be considered anything more than tentative. It ought not to be adopted until it has been tried thoroughly to see

whether it has in it the elements of permanency. It is novel to say the least to find numerous genera established on purely theoretical grounds with no known forms to put into them. In Dr. Fischer's classification the bacteria are divided into two orders: The Haplobacteriaceæ, or single celled bacteria, and the Trichobacteriaceæ, or thread-form bacteria (*Cladothrix*, etc.). The former multiply by slight elongation and cross-septation, the cells separating or remaining attached in small numbers. The latter consist of long cells, branched or unbranched, which finally break up into conidia or motile segments. The Haplobacteriaceæ consist of Coccaceæ, Bacillaceæ, and Spirillaceæ. The author's classification of the more difficult group is as follows:

FAMILY BACILLACEÆ.

Vegetative body one-celled, straight, with a distinct longitudinal axis, varying from short ellipsoidal to elongated rod form. Division always at right angles to the longitudinal axis; motile or non-motile; occurring singly or in chains; bearing endospores or arthrospores.

1. SUB-FAMILY BACILLEI.

Non-motile, destitute of flagella.

(a) With endospores.

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|---------------------------------------|---------------------------------|
| (1). <i>Bacillus</i> (Cohn). | Spore-bearing rods cylindrical. |
| (2). <i>Paracloster</i> (nov. gen.)* | Spore-bearing rods fusiform. |
| (3). <i>Paraplectrum</i> (nov. gen.)* | Spore-bearing rods clavate. |

(b. Without endospores, with arthrospores).

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|--------------------------------------|--|
| (4). <i>Arthrobacter</i> (De Bary).* | |
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2. SUB-FAMILY BACTRINEI.

Motile, with a single polar flagellum.

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|---|---------------------------------|
| (1). <i>Bactrinium</i> (nov. gen.) | Spore-bearing rods cylindrical. |
| (2). <i>Clostrinium</i> (nov. gen.)* | Spore-bearing rods fusiform. |
| (3). <i>Plectrinium</i> (nov. gen.)*? | Spore-bearing rods clavate. |
| (4). <i>Arthrobactrinium</i> (nov. gen.)* | With arthrospores. |
| (5). <i>Chromatium</i> . | Red sulphur bacteria. |

3. SUB-FAMILY BACTRILLEI.

Motile rods with a tuft of polar flagella.

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|--------------------------------------|---------------------------------|
| (1). <i>Bactrillum</i> (nov. gen.) | Spore-bearing rods cylindrical. |
| (2). <i>Clostrillum</i> (nov. gen.)* | Spore-bearing rods fusiform. |

- (3). *Plectrillum* (nov. gen.)* Spore-bearing rods clavate.
 (4). *Arthrobactrillum* (nov. gen.)* With arthrospores.

SUB-FAMILY BACTRIDEI.

Motile, with diffuse flagella.

- (1). *Bactridium* (nov. gen.) Spore-bearing rods cylindrical.
 (2). *Clostridium* (Prazm. *pro. parte.*) Spore-bearing rods fusiform.
 (3). *Plectridium* (nov. gen.) Spore-bearing rods clavate.
 (4). *Diplectridium* (nov. gen.) Spore-bearing rods dumb-bell shape.
 (5). *Arthrobactridium* (nov. gen.)* With arthrospores.

According to the author, 8 or nearly one-half of these so-called genera are founded on purely theoretical considerations, while there is some doubt as to whether there are any known species to go into two others. These pseudogenera are here indicated by asterisks.

—ERWIN F. SMITH.

The Mushroom Gardens of South American Ants.—Ever since the appearance of that wonderfully interesting book, *The Naturalist in Nicaragua*, it has seemed probable that the leaf-cutting ants do actually grow fungi for food, and use the countless thousands of leaf fragments which they drag into their nests for the same purpose that a gardener uses dung. Belt ascertained that the leaves were never used for food, found the fungus in every nest, observed the solicitude of the ants when it was disturbed, and in various particulars carried his inquiry as far as it was possible to do by simple observation. It remained for Alfred Möller, a young German, the nephew of Dr. Fritz Müller, and the pupil of Dr. Oscar Brefeld, not only to confirm Belt's surmise by close observation and exact experiment, but also to add greatly to our knowledge of the habits of these curious little gardeners and of the nature of the fungi they cultivate. These observations and experiments are embodied in *Die Pilzgärten einiger südamerikanischer Ameisen* (pp. VI, 127, Figs. 4, Pl. VII), which forms the 6th part of Professor Schimper's *Botanischen Mittheilungen aus den Tropen*, Jena, 1893. Möller's observations were made at Blumenau, Brazil, where he remained two years. The journey was made under the auspices of the Royal Academy of Sciences, of Berlin, whose wisdom in making this expenditure of a few thousand marks has certainly been more than justified by the outcome. During the course of the investigation several hundred ant nests were examined, these ants belonging to three genera, viz.: *Atta* (4 sp.); *Apterostigma* (3 sp.), and *Cyphomyrmex* (2 sp.) All are zealous cultivators and eaters of fungi, but the ants of

each genus grow a different sort, one kind only, and stubbornly refuse to eat any other, preferring to starve. More curious still, under the zealous attention of these little gardeners a special form of the fungus has been developed in much the same way that human selection has developed choice cabbages and cauliflowers out of what were originally quite ordinary sorts. This form of the fungus consists of groups of swollen hyphæ-ends, called Kohlrabi tufts. The greater part of the book deals with the fungous gardens of species of the genus *Atta*. The garden occupies the center of each nest as a loose, sponge-like mass, consisting of leaf-fragments held together by fungous threads. These gardens are often of large size, but between them and the walls of the nest there is always an open space. In the sponge-like cavities of these gardens the ants live, place their eggs, and rear their young. Often the eggs and sometimes the larvæ are overgrown and fastened together by the fungus, so that many as a hundred eggs may be seized and carried away by a single ant without inconvenience. The well known care that ants bestow on their progeny makes it certain that this placing the eggs in groups and allowing them to be bound together by the fungus is not simply accidental. When the nest is broken open and its contents scattered, or when the colony migrates, every tiny fragment of the fungous garden is gathered up and removed as carefully, and with as much solicitude as are the young. These fragments are rapidly and skillfully built into a new garden in the old nest or in some other place. Leaves are cut from a great many sorts of plants and often in such quantities as to entirely defoliate them, but are never eaten even to prevent starvation. Their sole food is the fungus which they cultivate, even fruits and starchy foods being used exclusively as a substratum for growing this much-beloved fungus. The leaf fragments brought into the nest are bitten and trimmed into smaller pieces and these are squeezed and kneaded into tiny pellets which are then carefully patted into the walls of the garden, and are overgrown by the fungus in a few hours. Exhausted fragments are thrown out and fresh pellets put in wherever needed by the fungus. Old worn-out masses of mycelium are also thrown out of the nest. Upon a special class of the colony, distinguished from the leaf cutters by their smaller size, devolves the task of weeding the garden and keeping it pruned within bounds. When neglected for a single day, i. e., by the removal of most or all of the ants, innumerable fungous threads shoot out into the air in every direction, and the well-kept garden soon becomes an unmanagable and uninhabitable thicket. When only a few ants are left in such a nest they work desparately, night and day, to keep it in order, but seem to know

that something is wrong, and are finally driven out by the too luxuriant growth of their own culture plant, being compelled to seize their young and flee for very life in a comical way. Most remarkable of all, especially to one who has busied himself much with trying to make and keep pure cultures of various fungi, is the ability of these ants to keep their gardens free from bacteria and all sorts of intruding fungi. Cultures made from various parts of a great many gardens showed conclusively that in an overwhelming proportion of cases these gardens are pure cultures of a single fungus. Unquestionably the ants must be constantly busy with the destruction and removal of intruding organisms. The Kohlrabi, or specially developed bunches of swollen hyphæ ends, occur as minute glistening rounded specks on all parts of the garden and are eagerly devoured by the ants. Unswollen, long mycelial threads push out into the air from all parts of the garden as soon as the ants are removed, and finally bear two kinds of conidial fruits, but nothing of the sort occurs while the ants are in undisturbed possession, and it is pretty certain that they must keep these undesirable shoots in check by constant biting, although this was not observed. The two kinds of conidial fruits were also obtained from artificial cultures under special conditions. In rare cases (only 4 were observed) the fungous garden pushes up through the top of the nest and fruits in the open air, this form of fructification being a large, flecked, wine-red, Amanita-like Agaricus, named by the author *Rozites gongylophora*, and never found except on the ant nests, rooted in the fungous garden. Pure cultures in great numbers and numerous microscopic observations proved beyond reasonable doubt that the swollen hyphæ, and the various kinds of fructification belong to one and the same fungus, and establish for the first time the existence of true conidia in the Agaricineæ. The ants of the other two genera, while equally diligent cultivators of fungi, build much smaller nests and are not leaf cutters, but use fragments of wood, dung, etc., as a substratum for their gardens. The fungi cultivated by them are believed to be hymenomycetous, but each genus has a different species. The different species of these ants vary in ability as gardeners. The facts set forth in this book were derived from prolonged examination of the ants in the open and in captivity, and by hundreds of patient and painstaking cultures and microscopic studies, and appear to be worthy of full credence. Mr. Möller's persistent and painstaking method of work is especially commendable to those over-ambitious young men who are content to look into the microscope one day and publish the next.

NOTE. Since this was written Mr. W. T. Swingle has discovered that our own *Atta tardigrada* has the same habits as its South American relatives. Several fungous gardens have been taken from nests near Washington, and the writer has seen beautiful Kohlrobi tufts growing on the dung of leaf-eating insects. ERWIN F. SMITH.

ZOOLOGY.

Irish Fresh-Water Sponges.—In a recent number of the *Irish Naturalist* (Vol. iv, pp. 122–131), Dr. R. Hanitsch enumerates six species of Spongillidæ from Ireland, the “British fauna” containing but four species. Three of these occur in Ireland, the other three sponges, all from the west coast of the latter country, being also North American species. Dr. Hanitsch would not solve this interesting distributional problem by supposing a former extension of the sponges over the whole northern hemisphere; he believes that their gemmules could readily have been carried across the Atlantic by winds, ocean currents, or birds. In some remarks on the European distribution of the Spongillidæ, Dr. Hanitsch notices their extreme rarity in southern Europe. Only one species is known from the Iberian peninsula (N. Portugal), two from the Italian, while none at all have been found in the Balkan. (Natural Science, July, 1895.)

Reproduction of the Edible Crab.—Through the observations of Mr. Gregg Wilson, some new facts have been brought to light concerning reproduction in the edible crab (*Cancer pagurus*) of the Northumberland coast, England. Crabs that have recently cast their shells have pale ovaries that show no development of ova to the naked eye. Hard crabs have brilliant orange or scarlet ovaries, with ova distinctly visible. Both lots are taken in the catch from October to February. Spawning seems to take place only every second year of the crab's life. At no time were ova undergoing segmentation found within the crab, so that the old idea that fertilization is internal must be abandoned. Milt is undoubtedly passed by the male crab into the body of the female, but it does not affect the roe before extrusion. It is received in flask-shaped *receptacula seminis*, that open off the oviducts quite near the genital apertures. They are well-valved and seem to retain the motionless spermatozoa for long periods. Spawning was noticed to